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(54) Preparation of food-grade edible oils

(57) Marine oil is stabilized by treatment with silica in the presence or absence of carbon, vacuum steam deodorisation at a temperature between about 140° C and about 210° C in the presence of 0.1-0.4% deodorised rosemary or sage extract. If desired 0.01-0.03% ascorbyl palmitate and 0.05-0.2% mixed tocopherol can be added.

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Description

[0001] The present invention relates to the preparation and stabilisation of food-grade marine oils.

[0002] Marine oils have attracted substantial interest as a source of n-3 long-chain polyunsaturated fatty acids (LCPUFA), particularly eicosapentaenoic acid (EPA) and docohexaenoic acid (DHA), which are of dietary significance. These LCPUFA contain 5 or 6 double bonds which render them prone to atmospheric oxidation accompagnied by a fishy taste and smell. The increasing interest in LCPUFA has prompted a research into methods of stabilizing fish oils against oxidation and off-flavor development.

[0003] It has been known for a long time that refined marine oils are initially free from a taste and smell of fish but that reversion through oxidation occurs rapidly. Many attempts have been made to stabilize the oils by the addition of different anti-oxidants or mixtures thereof. However, all these attempts failed so far, cf. R.J. Hamilton et al., Journal of American Oil and Chemist's Society (JAOCS), Vol. 75, no. 7, p. 813-822, (1998). Accordingly, there has been and still is a need for a process by means of which such marine oils can be stabilized over a long period of time in a simple and economical way whereby even after a long period of storage no fishy taste and smell occur.

[0004] Refined marine oil which has been treated with silica and stabilised with a mixture of lecithin, ascorbyl palmitate and alpha tocopherol in accordance with the procedure described in European Patent Publication 612 346 shows excellent rancimat stability and good application performance mainly for health food supplements. In dairy applications such as yoghurts and milk drinks, however, this oil develops a strong fish smell and taste.

[0005] Refined marine oil which has been treated with an adsorbent such as silica and stabilised with 0.1% deodorised rosemary extract (HERBALOX "O", Kalsec, Incorporated of Kalamazoo, Michigan) and, respectively, sage extract in accordance with and, respectively, an analogous manner to the procedure described in European Patent Publication 340 635 has a herby taste and smell which can be detected in food applications. This herby taste and smell supresses the taste and smell of fish. In dairy applications, the use of as little as 0.03% of HERBALOX "O" and, respectively, sage extract in the marine oil results in a very strong herby taste and smell which prevents the use of this oil in these applications.

[0006] It has now surprisingly been found in accordance with the present invention that marine oil which has been treated with silica in accordance with the procedure described in European Patent Publication 612 346 can be stabilized over a long period of time without the occurrence of fishy taste and smell by vacuum steam deodorization at a temperature between about 140° C and about 210° C in the presence of 0.1-0.4% of deodorised rosmary or sage extract.

[0007] The fully refined marine oil used in the present invention is one which has been neutralised, bleached and deodorised in a conventional manner. The oil can be, for example, menhaden oil, herring oil, sardine oil, anchovy oil, pilchard oil, tuna oil, hake oil etc. or a blend of one or more of these oils.

[0008] Factors associated with or even responsible for the fishy taste and smell of a marine oil are not-well defined. In order to get more information which factors are responsible for the fishy taste and smell, 21 oil samples were analyzed in detail as shown and discussed below. Samples 1-10 used in these analytical proceedings are commercially available standard fish oils from suppliers throughout the world which are regarded as "aged" because of the delays in refining them once more in accordance with the procedure described in European Patent Publication 612 346, whereas samples 11-15 are refined fish oils where it is known that both the extraction and refining have been done immediately after the fish have been caught or with mimimum delay only. Samples 16-17 are oils of fungal origin. Samples 18-21 have been produced from commercially available fish oils in accordance with the procedure described in European Patent Publication 612 346 in which, however, a special short path distillation step has been included at the start of the process to trap smell molecules for use as described below. The purpose to this wide trawl is to have as representative a range as possible of refined oils containing EPA and DHA.

[0009] Table 1 records the influence of the acid value, the EPA and, respectively, DHA content, the color and the pro-oxidant iron and copper levels on sensory responses of a trained panel to the above described 21 oil samples.

[0010] The analysis for the determination of the EPA and DHA content and, respectively, the pro-oxidant iron and copper levels were performed according to analytical methods known in the art. For determining the acid value, i.e. the number of milligrams of potassium hydroxide required to neutralize the free fatty acids in 1 gram of oil, the oil sample is titrated with 0.1N aqueous potassium hydroxide solution using a 1% phenolphthalein indicator. The size of the sample was determined as folllows:

Expected acid value	Test sample (g)
>0.5	40
0.5 to 1	20

(continued)

Expected acid value	Test sample (g)
1 to 5	5
5 to 10	2.5
10 to 20	1
>20	0.5

[0011] The color is determined by means of a Lovibond tintometer Model E AF 900 by matching the color of light transmitted through a specified depth of oil to the color of the lght originating from the same source, transmitted through standard color slides. The results are expressed in terms of the red (R), yellow(Y) and blue (B) units to obtain the match and the size of the cell used. Taste and smell are sensorically evaluated by a trained panel comprising 12-15 persons. The panelist are asked to rank the samples in terms of perceiption of fishy taste and smell. A hedonic scale of 1 to 5 is used to express the extent of fishiness in which 1 represents no fishy taste or smell, while 5 stands for a very strong fishy taste or smell. The samples are coded using a three-digit code and 10 - 15 ml are submitted to the panel in a plastic beaker at 22°C. The products are evaluated after processing and after 4 weeks and, respectively, 12 weeks storage at 22° C in aluminium containers.

[0012] Table 2 shows the effect of primary and secondary oxidation levels on the taste and smell of the same marine oils as in Table 1. Primary oxidation is measured as the peroxide value of the oils in milliequivalent (meq)/kg of oil. Secondary oxidation is measured in two ways: first by the reaction of unsaturated aldehydes in the oil with anisidine and by the reaction of alkanals, alkenals and alkadienals in the oil with N,N-dimethyl-p-phenylenediamine.

[0013] For determining the peroxide value the oil is treated in a solution of acetic acid and chloroform with a solution of iodide and subsequently the free iodine is titrated with a solution of sodium thiosulphate. The size of the sample was determined as follows:

Expected peroxide value	Test sample (g)
<1	10
1 to 5	2
5 to 10	1
>10	0.5

[0014] The p-anisidine value is defined as 100 times the absorbence measured at 350 nm in a 1 cm cell of a solution containing 1.0g of the oil in 100 ml of a mixture of hexane and a solution of p-anisidine in glacial acetic acid (0.025g/100 ml of glacial acetic acid). The size of the sample was determined as follows:

Expected p-anisidine value	Test sample (g)
0-5	5
5-10	3
10-20	2
20-30	1

[0015] The aldehyde values werde determined based on a method described by K. Miyashita et al., JAOCS, Vol. 68 (1991), according to which N,N-dimethyl-p-phenylenediamine is reacted with aldehydes in the presence of acetic acid. The three aldehyde classes (alkanal, alkenal and alkadienal) are determined by visible absorption at 400, 460

and, respectively, 500 nm. The aldehyde values are expressed in mmole/kg.

[0016] Furthermore, the level of smell molecules in each of these oils has been measured by static headspace coupled to GC/MS. The oil to be measured (samples of 1g each) is crimp sealed into a headspace vial (22ml) in an nitrogen atmosphere and heated at 120° C for 15 minutes in a headspace autosampler. A measured volume of the headspace is automatically injected onto a GC/MS using a heated transfer line. The gas chromatograph is used to separate the molecules and the mass spectrometer is used to identify and quantify the separated molecules. The results obtained are shown in Table 3.

Table 1

	Acid Value	EPA	DHA	Colour	Copper	Iron	Taste	Smell
		(%)	(%)		(ppb)	(ppb)		
Standard fish								
1	0.07	17.4	10.1	3.5R 23Y	13	39	2.3	0.7
2	0.06	18.8	9.1	1.1R 20Y	9	10	3.2	1.5
3	0.02	15.7	6.3	2.4R 24Y	6	16	2.8	1.2
4	0.04	11.6	12.1	2.6R 31Y	12	22	4.0	3.0
5	0.17	17.6	10.3	2.5R 20Y	17	24	2.1	0.8
6	0.08	16.9	11.7	3.1R30Y	31	29	2.8	1.6
7	0.04	6.7	27.7	1.6R 20Y	14	25	1.2	0.6
8	0.20	6.7	27.5	3.6R 32Y	37	9	1.2	0.5
9	0.04	6.6	27.3	1.5R 23Y	13	18	2.7	1.6
10	0.08	6.7	28.0	1.2R 31Y	12	12	3.6	2.0
Fresh fish								
11	0.32	6.9	13.0	0.8R 15Y	3	27	2.2	0.6
12	0.30	8.7	7.5	2.0R 25Y	7	24	2.4	0.8
13	0.20	11.8	13.3	1.6R 20Y	6	13	1.5	1.0
14	0.23	10.3	11.8	0.5R 5.4Y	8	26	2.6	1.3
15	0.23	8.6	12.6	1.5R 15Y	6	29	2.8	1.0
Single cell								
16	0.02	2.3	36.9	1.5R 32Y	7	10	1.7	0.9
17	0.77	0.4	31.0	1.2R 14Y	22	34	2.5	0.9
Standard fish distilled								
18	0.2	18.0	10.5	2.2R 20Y	3	24	0.7	0.7
19	0.23	18.0	10.4	2.2R 20 Y	5	24	0.6	0.7
20	0.22	18.1	10.5	2.2R 23Y	8	30	0.6	0.6
21	0.19	17.9	10.4	2.3R 22Y	7	24	1.0	0.5

[0017] The above table shows that there is no correlation between the acid value, the EPA and, repectively DHA content, the color and pro-oxidant iron and copper levels and the taste and smell of these marine oils.

Table 2

Description	Primary oxidation	Secondary oxidation					
	Peroxide value	p-Anisidine value	Aldehydes			Taste	Smell
Standard fish			A	B	C		
1	0.4	19.8	2.41	0.3	0.9	2.3	0.7
2	0.5	12.62	1.46	0.15	0.54	3.2	1.5
3	0.8	8.7	0.54	0.08	0.34	2.8	1.2
4	0.7	15.31	1.87	0.29	0.68	4.0	3.0
5	0	3.77	0.6	0.09	0.17	2.1	0.8
6	0	4.24	1.02	0.11	0.23	2.8	1.6
7	0.4	8.24	1.94	0.24	0.66	1.2	0.8
8	0.4	6.81	1.09	0.15	0.35	1.2	0.5
9	0.5	6.81	1.06	0.14	0.32	2.7	1.6
10	2.1	9.42	0.97	0.18	0.36	3.6	2.0
Fresh fish							
11	0	0.46	0.35	0.04	0.03	2.2	0.6
12	0	1.58	2.6	0.05	0.06	2.4	0.8
13	0	1.17	0.08	0.03	0.04	1.5	1.0
14	0	1.19	0.16	0.02	0.04	2.6	1.3
15	0	0.6	0.09	0.02	0.03	2.8	1.0
Single cell							
16	4	6.58	1.13	0.26	0.3	1.7	0.9
17	0	1.45	0.45	0.03	0.04	2.5	0.9
Standard fish distilled							
18	0	6.12	0.9	0.14	0.27	0.7	0.7
19	0	4.96	0.85	0.12	0.25	0.6	0.7
20	0	4.84	0.83	0.12	0.25	0.6	0.6
21	0	5.04	0.76	0.12	0.24	1.0	0.5

A = alkanals,
B = alkenals,
C = alkadienals

[0018] The above results show again that these oxidation indicators are not capable of distinguishing oils with a good taste and smell from those with bad ones.

Table 3

Description	Propanal	Propenal	Butanal	Ethyl Furan	Pentanal	Penten-3-one	Hexanal	Penten-3-ol	Heptanal	Hexenal	Octanal	Heptenal	Nonanal	Hexadienal	Octenal	Heptadienal	Taste	Smell
Standard fish																		
1	104	550	<40	11	<460	<90	590	218	<100	<470	<470	<490	<910	<960	<940	791	2.3	0.7
2	<90	371	<40	12	<460	<90	578	154	<100	<470	<470	<490	<910	<960	<940	757	3.2	1.5
3	214	840	<40	28	<460	<90	425	407	<100	<470	<470	<490	<910	<960	<940	701	2.8	1.2
4	516	1587	62	41	<460	151	<100	572	<100	<470	<470	<490	<910	<960	<940	<500	4.0	3.0
5	134	356	<40	<10	<460	405	<100	189	<100	<470	<470	<490	<910	<960	<940	<500	2.1	0.8
6	<90	280	<40	<10	<460	<90	<100	<90	<100	<470	<470	<490	<910	<960	<940	<500	2.8	1.6
7	599	4059	<40	91	<460	425	<100	<90	<100	<470	<470	1340.0	<910	1400	<940	<500	1.2	0.8
8	967	934	61	79	<460	175	<100	359	<100	<470	<470	<490	<910	<960	<940	<500	1.2	0.5
9	566	3877	41	85	<460	405	<100	1501	<100	<470	<470	<490	<910	<960	<940	<500	2.7	1.6
10	668	3430	79	58	<460	452	<100	1610	<100	<470	<470	<490	<910	<960	<940	1649	3.6	2.0
Fresh fish																		
11	<90	ND	<40	<10	<460	<90	<100	<90	<100	<470	<470	<490	<910	<960	<940	<500	2.2	0.6
12	109	ND	<40	15	<460	<90	<100	<90	<100	<470	<470	<490	<910	<960	<940	<500	2.4	0.8
13	<90	ND	<40	<10	<460	<90	<100	<90	<100	<470	<470	<490	<910	<960	<940	<500	1.5	1.0
14	<90	ND	<40	<10	<460	<90	<100	<90	<100	<470	<470	<490	<910	<960	<940	<500	2.6	1.3
15	<90	ND	<40	<10	<460	<90	<100	<90	<100	<470	<470	<490	<910	<960	<940	<500	2.8	1.0
Single cell																		
16	1992	1587	62	41	<460	151	<100	572	<100	<470	<470	<490	<910	<960	<940	<500	1.7	0.9
17	296	ND	<40	35	<460	293	231	261	<100	<470	<470	<490	<910	<960	<940	<500	2.5	0.9
Standard fish distilled																		
18	<90	122	<40	<10	<460	<90	902	91	<100	<470	<470	<490	<910	<960	<940	<500	0.7	0.7
19	<90	190	<40	<10	<460	<90	1319	92	<100	<470	<470	<490	<910	<960	<940	764	0.6	0.7
20	<90	170	<40	<10	<460	<90	1328	<90	<100	<470	<470	<490	<910	<960	<940	764	0.6	0.6
21	<90	203	<40	<10	<460	<90	1303	91	<100	<470	<470	<490	<910	<960	<940	833	1.0	0.5

[0019] Again, the above data show that static headspace cannot distinguish between good and bad tasting marine oils.

[0020] Tables 1 - 3 also show that marine oils which have been refined very soon after the oil has been extracted

from freshly caught fish do not show better sensory response than oils which have been refined from aged crude fish oil. However, levels of secondary anisidine reactives and aldehydes are extremely low in these fresh oils. These results suggest that whatever is responsible in the marine oil for the fishy taste and smell is present at extremely low levels below the detection limits of static headspace GC/MS. The data also show that neither anisidine nor aldehyde measurements are very useful in predicting the sensory quality of the oil - they are too insensitive.

[0021] Tables 1 - 3 show sensory data for single cell oils which demonstrate that they too can become fishy in both taste and smell. Table 1 also shows that when using specially refined oils it is possible to produce marine oils with excellent taste and smell but with quality parameters such as anisidine, peroxide, iron, copper, color and static headspace values which are not different from those of oils with poor taste and smell.

[0022] In order to have some understanding of the extent of the problem of the occurrence of fishy taste and smell in marine oils, efforts have been made to try and identify and quantify the molecules being responsible for the fishy taste and smell. Marine oils (1 kg each) rich in EPA and/or DHA which had a strong fishy smell were passed slowly through a short path still at 120° C and under reduced pressure (0.005 mbar). Two vacuum traps were connected in series each cooled with liquid nitrogen to collect the fishy volatiles which were removed by this process. These oils were then deodorised at 190° C and are the four specially refined oils recorded in Table 1 -3 as samples 18 - 21. Even though their traditional quality parameters are not different from those oils which were deemed fishy they had only little or no fishy taste. The condensates in the vacuum traps were dissolved in methyl tertiary butyl ether and subjected to olfactory detector GC/MS to identify fishy molecules which had been removed by this process. According to olfactory detector GC/MS the outlet stream from a gas chromatograph is split and routed to two different detectors. In the present case, the detectors used were the mass spectrometer and the human nose. Such a system allows peaks to be identified by the MS and assigned smell comments by an operator.

[0023] A number of very potent smell molecules were identified in the distillates and are recorded in Table 4.

Table 4

Target molecule	Characteristic according to prior art
4-heptenal	Fish oil
1-octen-3-one	Mushroom
1,5(Z)-octadien-3-ol	Mushroom
1,5(Z)-octadien-3-one	Metallic/fresh fish
(E,E)-2,4-heptadienal	Oxidized oil
(E)-2-octenal	Oxidized oil
(Z)-6-nonenal	Oxidized oil/putty/linseed oil
(E,Z)-2,6-nonadienal	Cucumber/fresh fish
(E)-2-nonenal	Oxidized oil
(E,Z)-1,3,5-undecatriene	Cod liver oil
(E,E)-2,4-decadienal	Fish/oxidized oil

[0024] As can be seen from Table 3, only a few of the above molecules could be identified using static headspace and, thus, a more sensitive method was needed to remove headspace molecules from the oils. The detection limits for e.g. 2-octenal and, respectively, 2,4-hexadienal were 940 ppb and, respectively, 500 ppb. In order to improve the sensitivity of detection, the technique of dynamic headspace has been used. According to this technique, 2 g aliquots of oil have been heated to 75° C in a water bath purged with helium (150 ml/min) through a Tekmar purge glass apparatus onto Perkin Elmer cartridges containing TENAX adsorbent (Enka Research Institute, Arnheim). The dynamic headspace has been measured by GC/MS using a 30m column of DB5-MS (1μm film thickness).

[0025] Table 5 shows the taste panel response to a number of blends of mixtures of marine oils and the dynamic headspace profile of a number of molecules. They have been identified by GC/MS and olfactory detector GC/MS. As can be seen, some of these molecules can be detected to single figure ppb level using dynamic headspace. The importance of the data in Table 5 is that they explain why the data in Table 1 - 3 cannot possibly correlate with marine oil taste and smell and they also demonstrate the very small amount of oxidation which is required before the oil deteriorates to a unacceptable quality from the point of its taste and smell.

Table 5

Sample No.	oil type	Taste	Concentration (ppb) 2,6-nonadienal	1,5-octdien-3-one	4-heptenal	2-hexenal	3,6-nonadienal	2,4-heptadienal	Taste factor
1	EPA	strong fish	53	69	26	146	457	213	4
2	EPA	middle fish	37	27	33	100	288	168	3
3	EPA	strong fish	68	54	53	232	710	542	4
4	EPA	no fish	28	151	35	113	254	282	1
5	EPA	no fish	19	74	31	79	223	208	1
6	EPA	middle fish	40	101	102	309	240	417	3
7	EPA	no fish	21	36	15	50	72	81	1
8	DHA	middle fish	27	79	61	230	324	257	3
9	DHA	no fish	23	64	15	57	101	128	1
10	EPA	no fish	16	42	10	0	70	92	1
11	DHA	slight fish	34	123	56	313	182	277	2
12	EPA	no fish	18	96	13	36	128	168	1
13	DHA	no fish	16	52	9	40	104	107	1
14	DHA	no fish	17	41	16	40	61	75	1
15	DHA	slight fish	25	69	26	104	156	198	2
16	EPA	middle fish	25	64	23	89	321	212	3
17	EPA	no fish	20	54	13	54	70	111	1
18	EPA	middle fish	28	113	41	147	441	334	3
19	EPA	no fish	21	45	7	39	96	182	1
20	EPA	slight fish	22	109	13	64	305	240	2
21	EPA	no fish	13	80	8	38	194	158	1
22	DHA	middle fish	22	157	31	101	719	563	3
23	DHA	no fish	15	78	6	48	216	215	1
24	EPA	no fish	14	55	7	38	109	116	1
25	DHA	no fish	0	52	8	25	78	70	1
26	DHA	slight fish	39	111	12	41	239	201	2
27	DHA	no fish	0	0	4	9	63	56	1
28	DHA	no fish	18	59	11	38	159	153	1
29	EPA	very strong fish	646	587	1135	4105	5015	3690	5
30	DHA	no fish	0	0	13	40	105	111	1
31	DHA	slight fish	20	0	9	33	84	61	2
32	DHA	no fish	0	71	7	32	164	129	1
33	DHA	no fish	0	69	11	39	119	91	1
34	DHA	no fish	0	76	7	51	177	123	1
35	DHA	no fish	0	98	2	42	137	105	1
36	DHA	no fish	13	34	6	32	44	34	1

[0026] Table 6 shows the excellent agreement between the level of 6 specially selected molecules in the headspace of the oils and the ranking by the taste panel by using a multiple discriminant analysis. Multiple discriminant analysis (MDA) is a statistical test used to determine whether a given classification of cases into groups is a likely one. It will report whether the group assignment of a case is true or false. The final data are presented in a table with rows and columns corresponding to actual and estimated group membership respectively. In the frame of the present invention the classification obtained from the sensorical evaluation by the taste panel was the taste factor. The MDA analysis was done through a statistical package called UNISTAT version 4.51.

Table 6

	Group 1	Group 2	Group 3	Group 4	Group 5
Group 1	22 100%	0 0%	0 0%	0 0%	0 0%
Group 2	0 0%	5 100%	0 0%	0 0%	0 0%
Group 3	0 0%	0 0%	6 100%	0 0%	0 0%
Group 4	0 0%	0 0%	0 0%	2 100%	0 0%
Group 5	0 0%	0 0%	0 0%	0 0%	1 100%
Group					
1	Not fishy				
2	Slight fishy				
3	Middle fishy				
4	Strong fishy				
5	V strong fishy				
Molecule	Retention Index	Comparison to standard retension time	Mass spectrum		
(E)-2-hexenal	861	Yes	Yes		
(Z)-4-heptenal	903	Yes	Yes		
1,5-(Z)-octadien-3-one	986	Yes	Yes		
(E,E)- 2,4 heptadienal	1004	Yes	Yes		
3,6-nonadienal	1109	No	Yes		
(E,Z)-2,6-nonadienal	1159	Yes	Yes		

[0027] The retention index of a compound is calculated from injections of C5-C15 saturated straight chain hydrocarbons under the same chromatographic conditions as the analysis of interest and is similar to its retention time in that the longer it is retained on a GC column the greater is its retention index/time. The use of the retention indices rather than retention times makes the information more rigorous and transferable although the retention indices are still dependant on the column phase and chromatographic conditions but minimise instrument dependant variables.

[0028] In order for a peak on a GC trace to be accepted as having a certain identity certain conditions must be met. The traditional one with GC is that it should have the same retention index/time as an authentic standard. Of the 6 molecules listed standards were obtained for 5 of them. Alternatively, mass spectra can be used as an additional tool to confirm peak identity.

[0029] Table 7 shows the effect of increasing concentration of deodorised rosemary extract on the rancimat stability of a marine oil by adding it after deodorisation.

Table 7

Deodorised HERBALOX "O" added	Rancimat Induction Time (100° C)
(%)	(hours)
0	1.70
0.25	3.02
0.5	3.87
0.75	4.93
1.0	5.45
1.5	5.73
2.0	6.98
2.5	7.65
3.0	8.23
3.5	9.28
4.0	10.7

[0030] Table 7 shows that between 0 and 4% addition of rosemary extract, the rancimat induction time and, thus, the rancimat stability of marine oil, increases with an increasing the amount of rosemary extract. Nevertheless, the use of rosemary extract as a stabiliser of marine oil in accordance with the prior art, i.e.after deodorisation, is - even at the low amount of 0.2% - disadvantageous due to the powerful herby smell of the commercial deodorised rosemary extract, particularly if it is put into dairy food applications. This makes it impossible to use the dose benefits shown in Table 7.

[0031] It has now surprisingly been found in accordance with the present invention that adding the rosemary extract to the oil before deodorisation removes the powerful smell without removing or destroying the anti-oxidant activity. The results of the relevant experiments are set forth in Tables 8 and 9.

[0032] Table 8 shows a range of headspace molecules which describe the headspace of deodorised rosemary extract at a concentration of 0.2% added to deodorised marine oil after deodorisation and, respectively, 0.2% and 0.4% added before deodorisation. In the latter case, two deodorisation temperatures are given.

Table 8

HERBALOX "O"	0.2%	0.4%	0.4%	0.2%	0.2%
Addition	After Deodorisation	Before deodorisation	Before deodorisation	Before deodorisation	Before deodorisation
Temperature	-	150° C	190° C	150° C	190° C
	% normalised/relative	% removed	% removed	% removed	% removed
Limonene	100 / 4.7	17	20	50	50
Eucalyptol	100 / 3.5	100	100	100	100
Linalool	100 / 1.5	100	100	100	100
Linalyl propanoate	100 / 3.8	100	100	100	100
Camphor	100 /20.3	97	99	100	100
Iso-Borneol	100 / 3.8	100	100	93	90
Fenchyl acetate	100 /27.3	100	100	100	100
Vebenone	100 / 3.0	100	100	100	100
Bornyl acetate	100 / 1.2	100	100	100	100
Copaene (1)	100 / 1.8	100	100	100	100
Ioscaryophyllene	100 / 0.6	20	20	20	20
Caryophyllene	100 /27.9	84.8	100	100	100
Copaene	100 / 0.5	100	100	100	100

[0033] The relative values given in column 2 were derived from the analysis of marine oil with 0.2% HERBALOX "O" added after deodorising. When the oils are deodorised it is necessary to have a concentration against which it is possible to measure removal of the headspace molecules. Therefore, the concentration of each compound found in the experiment in which the rosemary extract was added after removal was taken as 100% and the effects of deodorising measured against this level.

[0034] Table 8 shows that when a mineral oil to which 0.2% of rosemary oil was added before deodorisation is deodorised at 150° C or 190° C virtually all of these spicey molecules are removed from the oil. With 0.4% addition, removal of most of the spicey molecules is low, whereby particularly two of the main components, i.e. camphor and caryophyllene, are not completely removed.

[0035] The herby smell in an oil deodorised at 150° C with 0.4% addition of rosemary extract before deodorisation is still strong whereas an oil with only 0.2% rosemary extract added does not have any herby smell.

[0036] Table 9 shows the effect on the anti-oxidant system depending on the deodorisation temperature, anti-oxidant mixture and whether the rosemary is added before or after the deodorisation.

Table 9

Addition	HERBALOX "O"	Sage Extract	Ascorbyl Palmitate	Mixed Tocopherol	Deodorisation Temperature	Rancimat Induction Time
	(%)	(%)	(%)	(%)	(°C)	(hours)
-	-	-	-	-	-	1.7
After	0.2	-	-	-	-	3.0
After	0.2	-	-	-	150	3.0
After	0.2	-	-	-	190	2.9
Before	0.2	-	-	-	150	3.3

Table 9 (continued)

Addition	HERBALOX "O"	Sage Extract	Ascorbyl Palmitate	Mixed Tocopherol	Deodorisation Temperature	Rancimat Induction Time
	(%)	(%)	(%)	(%)	(°C)	(hours)
Before	0.2	-	-	-	190	4.1
Before	0.2	-	0.02	0.1	150	5.4
Before	0.2	-	0.02	0.1	190	6.2
After	-	0.2	-	-	190	2.3
Before	-	0.2	-	-	190	3.4
Before	-	0.2	0.02	0.1	190	5.3

[0037] Adding 0.2% rosemary extract to the marine oil without deodorising, increases the rancimat stability from 1.7 to 3.0 hours at 100° C. The same or about the same rancimat stability is seen when the rosemary extract is added to the oil after deodorising at 150° C and 190° C. An only slightly increased rancimat stability in seen when sage extract is added to the oil after deodorising at 190°C. If the rosemary extract is added to the oil before the deodorisation at 150° C there is a slightly increased rancimat stability but by deodorising at 190° C in the presence of rosmary and, respectively, sage extract the rancimat stability of the oil is increased substantially to 4.1 and, respectively, 3.4 hours. Addition of 0.02% ascorbyl palmitate and 0.1% mixed tocopherol after deodorisation further enhances the rancimat stability of the oil. Thus, by deodorising the oil at 190° C and adding 0.2% rosemary and, respectively, sage extract before the deodorisation followed by 0.02 % ascorbyl palmitate and 0.1% mixed tocopherol after the deodorisation it is possible to increase the rancimat stability of the oil from 1.7 to 6.2 and, respectively, 5.3 hours.

[0038] Accordingly, an object of the present invention is a process for the preparation and stabilizaion of food-grade marine oil by treating marine oil with silica in the presence or absence of carbon, vacuum steam deodorising at a temperature between about 140° C and about 210° C in the presence of 0.1-0.4% rosemary or sage extract and, if desired, adding 0.01-0.03% ascorbyl palmitate and 0.05-0.2% mixed tocopherol, as well as the use of the oil thus obtained in food applications. A further object of the present invention is a method of determining the sensory quality of a unknown marine oil by measuring the dynamic headspace profile of the marine oil with regard to the 6 following compounds:

(Z)-4-heptenal
(E)-2-hexenal
1,5-(Z)-octadien-3-one
(E,E)-2,4-heptadienal
3,6-nonadienal
(E,Z)-2,6-nonadienal

and evaluating the results abtained against the results of the oils given in Table 5 by multiple discriminant analysis.

[0039] Preferably, the silica treatment is performed in the presence of carbon. The preferred temperature for the deodorisation step lies between 150° C and 190° C, more preferred at about 190° C. The preferred amount of deodorised rosemary or sage extract present during deodorisation is 0.2%. Furthermore, it is preferred to add after deodorisation 0.01-0.03%, preferably 0.02%, ascorbyl palmitate and 0.05-0.2%, preferably 0.1%, mixed tocopherol.

[0040] The following examples illustrate the invention, but do not limit its scope in any manner. The silica and carbon used in the present invention has been described in detail in European Patent Publication 612 346. All oils used had been mixed with 5% silica and 2% activated carbon at 80° C and then filtered as described in European Patent Publication 612 346. The filtered product is called "adsorbed oil" in the examples.

Example 1

[0041] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were deodorised at 190° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. The oil was then divided into aliquots to have additions of HERBALOX "O" up to 4% and was used to provide the rancimat stabilities recorded in Table 7. To a separate aliquot of this oil, 0.2% HERBALOX "O" was added. Results of this study are recorded in Table 9. Samples of this oil were also dynamically purged to measure the content of spicey headspace molecules from the HERBALOX "O" " addition. These results are recorded in Table 8.

Example 2

[0042] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were deodorised at 150° C for 2 hours then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. 0.2% HERBALOX "O" was added to this oil. Results of this study are recorded in Table 9. Samples of this oil were also dynamically purged to measure the content of spicey head-space molecules from the HERBALOX "O" " addition. These results are recorded in Table 8.

Example 3

[0043] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were mixed with 0.2% HERBALOX "O", then deodorised at 190° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. The oil was then divided into aliquots to have no addition of further anti-oxidant and, respectively, addition of 0.02% ascorbyl palmitate and 0.1% mixed tocopherol. Rancimat stabilites are recorded in Table 9. Samples of this oil were also dynamically purged to measure the content of spicey headspace molecules from the HERBALOX "O" addition. The results are recorded in Table 8.

Example 4

[0044] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were mixed with 0.2% HERBALOX "O", then deodorised at 150° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. The oil was then divided into aliquots to have no addition of further anti-oxidant and, respectively, addition of 0.02% ascorbyl palmitate and 0.1% mixed tocopherol. Rancimat stabilites are recorded in Table 9. Samples of this oil were also dynamically purged to measure the content of spicey headspace molecules from the HERBALOX "O" addition. The results are recorded in Table 8.

Example 5

[0045] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were mixed with 0.4% HERBALOX "O", then deodorised at 150° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. The oil was then divided into aliquots which were dynamically purged to measure the content of spicey headspace molecules from the HERBALOX "O" addition. The results are recorded in Table 8.

Example 6

[0046] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were mixed with 0.4% HERBALOX "O", then deodorised at 190° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. The oil was then divided into aliquots which were dynamically purged to measure the content of spicey headspace molecules from the HERBALOX "O" addition. The results are recorded in Table 8.

Example 7

[0047] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were deodorised at 190° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. 0.2% sage extract was added to this oil. Results of this study are recorded in Table 9.

Example 8

[0048] 950 g of adsorbed marine oil containing 11.0% EPA and 17.8% DHA were mixed with 0.2% sage extract, then deodorised at 190° C for 2 hours and then cooled to 60° C. The steam was stopped and replaced by a nitrogen purge for 5 minutes. The oil was then divided into aliquots to have no addition of further anti-oxidant and, respectively, addition of 0.02% ascorbyl palmitate and 0.1% mixed tocopherol. Rancimat stabilites are recorded in Table 9.

[0049] The following examples illustrate the use of marine oil obtained in accordance with the present invention in practical food applications. The oil used is hake oil containing 11.0% EPA and 17.8% DHA which was deodorised at 190° C in the presence of 0.2% HERBALOX "O" and will be named in the examples as "ROPUFA '30' n-3 Food Oil".

Example 9

Soft Drink with 30% juice

[0050]

Typical serving: 300 ml
n-3 LCPUFA content: 75 mg/serving

	[g]
Part I	
Orange concentrate 60.3° Brix, 5.15% acidity	657.99
Lemon concentrate 43.5° Brix, 32.7% acidity	95.96
Orange flavour, water soluble	13.43
Apricot flavour, water soluble	6.71
Water	26.46
Part II	
β-Carotene 10% CWS	0.89
Water	67.65
Part III	
Ascorbic acid	4.11
Citric acid anhydrous	0.69
Water	43.18
Part IV	
Stabiliser	1.37
Sodium benzoate	2.74
Water	64.43
Part V	
Orange flavour, oil soluble	0.34
Orange oil distilled	0.34
ROPUFA '30' n-3 Food Oil	13.71
Bottling syrup	
Softdrink compound	74.50
Water	50.00
Sugar syrup 60° Brix	150.00

[0051] The bottling syrup was diluted with water to 1 l ready to drink beverage.

Part I: All ingredients were mixed together without incorporation of air.

Part II: β-Carotene was dissolved in water.

Part III: Ascorbic acid and citric acid were dissolved in water.

Part IV: Sodium benzoate was dissolved in water. The stabiliser was added under stirring and swollen for 1 hour.

Part V: All ingredients were mixed together.

[0052] All parts were mixed together before homogenisation using first a Turrax and then a high pressure homogenizer (p_1 = 200 bar, p_2 = 50 bar).

[0053] Instead of using sodium benzoate, the beverage may be pasteurised. The beverage may also be carbonised.

Example 10

5 cereal bread

[0054]

Typical serving: 100 g
n-3 LCPUFA content: 90 mg/serving

	[%]
5 cereal flour	100.00
Water	70.00
Yeast	4.00
Salt	2.00
ROPUFA '30' n-3 Food Oil	0.56

[0055] The yeast was dissolved in a part of the water. All ingredients including ROPUFA '30' n-3 Food Oil were mixed together to form a dough. Salt was added at the end of the kneading time. After fermentation, the dough was reworked and divided before a loaf was formed. Before baking, the surface of the loaf was brushed with water and sprinkled with flour.

Parameters:

[0056]

• Kneading:	
Spiral kneading system	4 min 1st gear
	5 min 2nd gear
Dough proofing:	60 min
Dough temperature:	22 - 24° C
Proofing time:	30 min
• Baking:	
Oven:	Dutch type oven
Baking temperature:	250/220° C
Baking time:	50 - 60 min

[0057] Estimated baking loss: 10 %.

Example 11

Table Margarine

60% fatarine

[0058]

Typical serving: 30 g
n-3 LCPUFA content: 225 mg/serving

	[%]
Fat phase:	
Sunflower oil	25.220
Mixture of hardened rapeseed, soy, coconut and palm fat	31.175
ROPUFA '30' n-3 Food Oil	3.000
Emulsifier	0.600
Beta-Carotene 30% FS	0.004
Butter flavour, oil soluble.	0.001
Water phase:	
Water	39.858
Salt	0.100
Citric Acid	0.042

Fat phase:

[0059] The fats were melted, but not exceeding 60° C. The oil was added and kept at the same temperature. Shortly before processing, the ROPUFA '30' n-3 Food Oil was added. Then all other oil soluble ingredients were added to the fat/oil mixture.

Water phase:

[0060] All water soluble ingredients were dissolved in water and pasteurised.
[0061] The water phase was added slowly to the oil phase (50° C) and mixed with a high shear mixer to form a homogeneous emulsion. The emulsion was crystallised in a margarine plant, equipped with a mutator, pinworker and resting tube. The margarine was filled into cups at 20° C and kept cool.

Example 12

Table Margarine

80% fat

[0062]

Typical serving: 30 g
n-3 LCPUFA content: 225 mg/serving

	[%]
Fat phase:	
Sunflower oil	30.850
Mixture of hardened rapeseed, soy, coconut and palm fat	45.800
ROPUFA '30' n-3 Food Oil	3.000
Emulsifier	0.250
Beta-Carotene 30% FS	0.008
Butter flavour, oil soluble.	0.090
Water phase:	
Water	19.910
Salt	0.100
Citric Acid	0.005
Butter flavour, water soluble.	0.005

Fat phase:

[0063] The fats were melted, but not exceeding 60° C. The oil was added and kept at the same temperature. Shortly before processing, the ROPUFA '30' n-3 Food Oil was added. Then all other oil soluble ingredients were added to the fat-oil mixture.

Water phase:

[0064] All water soluble ingredients were dissolved in water and pasteurised.
[0065] The water phase was added slowly to the oil phase (50° C) and mixed with a high shear mixer to form a homogeneous emulsion. The emulsion was crystallised in a margarine plant, equipped with a mutator, pinworker and resting tube. The margarine was filled into cups at 15° C and kept cool.

Example 13

Cookies

Type Mailänder

[0066]

Typical serving: 25 g
n-3 LCPUFA content: 62.5 mg/serving

	[g]
Wheat Flour, type 550	410.0
Sugar	205.0
Fat/Butter	195.9
ROPUFA '30' n-3 Food Oil	9.1
Whole egg (liquid)	180.0

(continued)

	[g]
Lemon Flavour	q.s.
Baking agent	q.s.

[0067] The ROPUFA '30' n-3 Food Oil was added to the melted fat. All other ingredients were added slowly under mixing to form a sweet short pastry.

[0068] Afterwards, the pastry was kept cool (4° C) for at least 2 hours before flattening the pastry to a thickness of approx. 5 mm. Pieces were cut out and brushed with egg yolk on the surface before baking.

• Baking:	
Oven:	fan oven
Baking temperature:	180° C
Baking time:	15 min

Example 14

Toast

[0069]

Typical serving: 100 g
n-3 LCPUFA content: 90 mg/serving

	[%]
Wheat Flour, type 550	100.00
Water	60.00
Yeast	5.00
Salt	2.00
Fat/Butter	9.43
ROPUFA '30' n-3 Food Oil	0.57
Malt	1.00
Emulsifier baking agent	2.50

[0070] The yeast was dissolved in a part of the water. All ingredients were mixed together to form a dough including ROPUFA '30' n-3 Food Oil. Salt was added at the end of the kneading time. Afterwards, the dough was reworked, divided and placed in a baking tin for fermentation. After baking, the loaf was unmoulded directly.

Parameters:

[0071]

• Kneading:	
Spiral kneading system	5 - 6 min 1st gear 3 - 4 min 2nd gear
Dough proofing:	none
Dough temperature:	22 - 24° C
Proofing time:	40 min
• Baking:	
Oven:	Dutch type oven
Baking temperature:	220° C
Baking time:	35 - 40 min

Example 15

Whole flour biscuits

[0072]

Typical serving: 25 g
n-3 LCPUFA content: 125 mg/serving

	[g]
Whole wheat flour	355.0
Fat	195.3
ROPUFA '30' n-3 Food Oil	18.2
Cane sugar	177.5
Almond, ground	118.0
Whole egg (liquid)	130.0
Salt	1.0
Baking agent	2.5
Cinnamon	2.5
Lemon Peel flavour	q.s.
Lemon Juice	q.s.

[0073] The ROPUFA '30' n-3 Food Oil was added to the melted fat. Then all other ingredients were added slowly under mixing to form a sweet short pastry.

[0074] Afterwards the pastry was kept cool (4° C) for at least 2 hours before flattening the pastry to a thickness of approx. 6 mm. Pieces were cut out and brushed with egg yolk on the surface and sprinkled with cane sugar before baking.

Parameters:

[0075]

• Baking:	
Oven:	fan oven
Baking temperature:	200° C
Baking time:	10 min

[0076] Estimated baking loss 10 %.

Example 16

Yoghurt cake

[0077]

Typical serving: 100 g
n-3 LCPUFA content: 250 mg/serving

	[g]
Wheat flour	310.0
Sugar incl. Vanilla sugar	240.0
Whole egg (liquid)	200.0
Yoghurt	170.0
Fat/Oil	60.9
Baking agent	10.0
ROPUFA '30' n-3 Food Oil	9.1

[0078] ROPUFA '30' n-3 Food Oil was added to the fat/oil. The yoghurt was mixed with sugar, vanilla sugar and eggs before the addition of the fat/oil containing ROPUFA '30' n-3 Food Oil, the flour and baking agent. The dough was beaten for at least 5 min. at medium speed. The batter was then spread into cake tins and baked in an oven.

Parameters:

[0079]

• Baking:	
Oven:	Fan oven
Baking temperature:	190° C
Baking time:	40 min

Example 17

UHT Milk Drink

1.7% fat

[0080]

Typical serving: 300 ml
n-3 LCPUFA content: 150 mg/serving

	[%]
Part I	
ROPUFA '30' n-3 Food Oil	0.200
Milk 1.5 % fat	2.580
Part II	
Part I	2.780
Sodium ascorbate	0.025
Milk 1.5 % fat	97.195

Pre-emulsion

[0081] Part I was mixed together and homogenised in high pressure homogenizer (p_1 = 150 bar, p_2 = 50 bar) to reach an homogeneous emulsion.

UHT-Procedure:

[0082] Part I was added together with sodium ascorbate to the rest of the milk without incorporation of air. The mix was homogenised in a high pressure homogenizer (p_1 = 150 bar, p_2 = 50 bar) and preheated in a tubular heat exchanger before thermal processing in a direct heat exchanger at 140° C for 4 sec, vacuum-cooling and aseptically packaging.

Example 18

Yoghurt - set type

3.5% fat

[0083]

Typical serving: 150 g
n-3 LCPUFA content: 225 mg/serving

	[%]
Full fat milk (3.8% fat)	75.0
Skimmed milk (0.1% fat)	14.9
Skimmed milk powder	2.0

(continued)

	[%]
Sugar	5.0
Yoghurt	2.5
ROPUFA '30' n-3 Food Oil	0.6

[0084] The milk was heated to 35° C before addition of milk powder and sugar. This mixture was heated to 65° C to dissolve all ingredients. ROPUFA '30' n-3 Food Oil was added to the mixture before the homogenisation in a high pressure homogenizer (p1 = 150 bar, p2 = 50 bar) at 65° C. This emulsion was then pasteurised at 80 °C for 20 minutes. After cooling to 45° C natural yoghurt/culture was added and mixed. Then this mixture was filled into cups and fermented at 45° C for 3-4 hours until a pH of 4.3 was reached and then stored at 4° C.

Example 19

Yoghurt - stirred type

3.5% fat

[0085]

Typical serving: 150 g
n-3 LCPUFA content: 225 mg/serving in yoghurt

	[%]
Full fat milk (3.8% fat)	78.8
Skimmed milk (0.1% fat)	10.8
Skimmed milk powder	2.0
Stabiliser	0.3
Sugar	5.0
Yoghurt	2.5
ROPUFA '30' n-3 Food Oil	0.6

[0086] The milk was heated to 35° C before addition of milk powder, stabiliser and sugar. This mixture was heated to 65° C to dissolve all ingredients before homogenisation in a high pressure homogenizer (p_1 = 150 bar, p_2 = 50 bar) at 65° C. This emulsion was then pasteurised at 80° C for 20 minutes. After cooling to 45° C natural yoghurt/culture was added and mixed, followed by a fermentation at 45° C for 3-4 hours until a pH of 4.3 was reached. After cooling and stirring vigorously, the yoghurt was filled in cups and stored at 4° C.

Method A:

[0087] Addition of ROPUFA '30' n-3 Food Oil before homogenisation.

Method B:

[0088] Addition of ROPUFA '30' n-3 Food Oil after fermentation while stirring.

Claims

1. A process for the preparation and stabilizaion of food-grade marine oil, which process comprises treating marine oil with silica in the presence or absence of carbon, vacuum steam deodorising at a temperature between about

140° C and about 210° C in the presence of 0.1-0.4% rosemary or sage extract and, if desired, adding 0.01-0.03% ascorbyl palmitate and 0.05-0.2% mixed tocopherol.

2. A process according to claim 1, wherein the vacuum steam deodorisation is performed in the presence of rosemary extract.

3. A process according to claim 1 or 2, wherein the silica treatment is performed in the presence of carbon.

4. A process according to any one of claims 1 to 3, wherein the temperature for the deodorisation step lies between 150° C and 190° C, preferably at about 190° C.

5. A process according to any one of claims 1 to 4, wherein the amount of deodorised rosemary extract present during deodorisation is 0.2%.

6. A process according to any one of claims 1 to 5, wherein 0.01-0.03%, preferably 0.02%, ascorbyl palmitate and 0.05-0.2%, preferably 0.1%, mixed tocopherol is added after deodorisation

7. Use of a marine oil obtained according to any one of claims 1 to 6 for the preparation of food applications.

8. A method of determining the sensory quality of a unknown marine oil by measuring the dynamic headspace profile of the marine oil with regard to the 6 following compounds:

(Z)-4-heptenal
(E)-2-hexenal
1,5-(Z)-octadien-3-one
(E,E)-2,4-heptadienal
3,6-nonadienal
(E,Z)-2,6-nonadienal

and evaluating the results obtained against the results of the oils given in Table 5 by multiple discriminant analysis.

European Patent Office

EUROPEAN SEARCH REPORT

Application Number
EP 99 12 1655

DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
A D	WO 93 10207 A (HOFFMANN LA ROCHE) 27 May 1993 (1993-05-27) * example 8 * * claims 1,10,15,27,33 * & EP 0 612 346 A	1,3,4,6,7	C11B3/10 C11B3/14 C11B5/00 A23D9/00 G01N33/03
D,A	EP 0 340 635 A (KABIVITRUM AB) 8 November 1989 (1989-11-08) * page 5, line 36 - line 54 * * page 13; table 2 * * claims 1,2,6-9,11 *	1,7	
A	C. KARAHADIAN ET AL.: "Evaluation of compounds contributing characterizing fishy flavors in fish oils" JOURNAL OF THE AMERICAN OIL CHEMISTS' SOCIETY., vol. 66, no. 7, 1989, pages 953-960, XP002129666 AMERICAN OIL CHEMISTS' SOCIETY. CHAMPAIGN., US ISSN: 0003-021X * the whole document *	8	TECHNICAL FIELDS SEARCHED (Int.Cl.7)
A	C. MILO ET AL.: "Changes in the odorants of boiled salmon and cod as affected by the storage of the raw material" JOURNAL OF AGRICULTURAL AND FOOD CHEMISTRY., vol. 44, no. 8, 1996, pages 2366-2371, XP002129667 AMERICAN CHEMICAL SOCIETY. WASHINGTON., US ISSN: 0021-8561 * the whole document * -/--	8	C11B A23D G01N

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
THE HAGUE	4 February 2000	Dekeirel, M

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
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A : technological background
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EUROPEAN SEARCH REPORT

Application Number
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DOCUMENTS CONSIDERED TO BE RELEVANT

Category	Citation of document with indication, where appropriate, of relevant passages	Relevant to claim	CLASSIFICATION OF THE APPLICATION (Int.Cl.7)
A	C. KARAHADIAN ET AL.: "Low temperature deodorizations of fish oils with volatile acidic and basic steam sources" JOURNAL OF THE AMERICAN OIL CHEMISTS' SOCIETY., vol. 67, no. 2, 1990, pages 85-91, XP002129668 AMERICAN OIL CHEMISTS' SOCIETY. CHAMPAIGN., US ISSN: 0003-021X * the whole document *	8	
A	US 5 006 281 A (RUBIN DAVID ET AL) 9 April 1991 (1991-04-09) * column 4, line 47 - line 64 * * column 6, line 4 - line 8 * * column 9, line 5 - line 21 *	1,7	
			TECHNICAL FIELDS SEARCHED (Int.Cl.7)

The present search report has been drawn up for all claims

Place of search	Date of completion of the search	Examiner
THE HAGUE	4 February 2000	Dekeirel, M

CATEGORY OF CITED DOCUMENTS

X : particularly relevant if taken alone
Y : particularly relevant if combined with another document of the same category
A : technological background
O : non-written disclosure
P : intermediate document

T : theory or principle underlying the invention
E : earlier patent document, but published on, or after the filing date
D : document cited in the application
L : document cited for other reasons

& : member of the same patent family, corresponding document

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ANNEX TO THE EUROPEAN SEARCH REPORT ON EUROPEAN PATENT APPLICATION NO. EP 99 12 1655

This annex lists the patent family members relating to the patent documents cited in the above-mentioned European search report.
The members are as contained in the European Patent Office EDP file on
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04-02-2000

Patent document cited in search report	Publication date	Patent family member(s)	Publication date
WO 9310207 A	27-05-1993	AT 123799 T	15-06-1995
		CA 2123595 A,C	27-05-1993
		DE 69202986 D	20-07-1995
		DE 69202986 T	02-11-1995
		DK 612346 T	07-08-1995
		EP 0612346 A	31-08-1994
		ES 2073936 T	16-08-1995
		FI 942254 A	13-05-1994
		JP 2956909 B	04-10-1999
		JP 7501355 T	09-02-1995
		NO 941808 A	13-05-1994
		US 5855944 A	05-01-1999
EP 0340635 A	08-11-1989	US 4874629 A	17-10-1989
		AT 141637 T	15-09-1996
		AU 625415 B	09-07-1992
		AU 3388289 A	02-11-1989
		CA 1335110 A	04-04-1995
		DE 68926977 D	26-09-1996
		DE 68926977 T	16-01-1997
		ES 2091754 T	16-11-1996
		GR 3021643 T	28-02-1997
		JP 2016195 A	19-01-1990
		SG 43233 A	17-10-1997
		US 5023100 A	11-06-1991
US 5006281 A	09-04-1991	NONE	

EPO FORM P0459

For more details about this annex : see Official Journal of the European Patent Office, No. 12/82